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PALMER & DODGE, LLP KATHLEEN M. WILLIAMS 111 HUNTINGTON AVENUE BOSTON, MA 02199			EXAMINER SWITZER, JULIET CAROLINE	
			ART UNIT 1634	PAPER NUMBER
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**Please find below and/or attached an Office communication concerning this application or proceeding.**

The time period for reply, if any, is set in the attached communication.

## Office Action Summary

**Application No.**

10/809,675

**Applicant(s)**

LIEW, CHOONG-CHIN

**Examiner**

Juliet C. Switzer

**Art Unit**

1634

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

### Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

### Status

- 1) ☒ Responsive to communication(s) filed on 10/2/07; 10/11/07.
- 2a) ☒ This action is **FINAL**. 2b) ☐ This action is non-final.
- 3) ☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

### Disposition of Claims

- 4) ☒ Claim(s) 50-53 and 56-61 is/are pending in the application.
- 4a) Of the above claim(s) \_\_\_\_\_ is/are withdrawn from consideration.
- 5) ☐ Claim(s) \_\_\_\_\_ is/are allowed.
- 6) ☒ Claim(s) 50-53 and 56-59 is/are rejected.
- 7) ☒ Claim(s) 60-61 is/are objected to.
- 8) ☐ Claim(s) \_\_\_\_\_ are subject to restriction and/or election requirement.

### Application Papers

- 9) ☐ The specification is objected to by the Examiner.
- 10) ☐ The drawing(s) filed on \_\_\_\_\_ is/are: a) ☐ accepted or b) ☐ objected to by the Examiner.  
Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).  
Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).
- 11) ☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

### Priority under 35 U.S.C. § 119

- 12) ☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).
- a) ☐ All b) ☐ Some \* c) ☐ None of:
- ☐ Certified copies of the priority documents have been received.
  - ☐ Certified copies of the priority documents have been received in Application No. \_\_\_\_\_.
  - ☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

\* See the attached detailed Office action for a list of the certified copies not received.

### Attachment(s)

- |   |   |
|---|---|
| 1) <input type="checkbox"/> Notice of References Cited (PTO-892)                    | 4) <input type="checkbox"/> Interview Summary (PTO-413)           |
| 2) <input type="checkbox"/> Notice of Draftperson's Patent Drawing Review (PTO-948) | Paper No(s)/Mail Date. _____                                      |
| 3) <input type="checkbox"/> Information Disclosure Statement(s) (PTO/SB/08)         | 5) <input type="checkbox"/> Notice of Informal Patent Application |
| Paper No(s)/Mail Date _____   | 6) <input type="checkbox"/> Other: _____                          |

Art Unit: 1634

### **DETAILED ACTION**

1. This action is written in response to applicant's correspondence received 10/2/07 and 10/11/07. Applicant's amendments and arguments have been thoroughly reviewed, but are not persuasive to place the claims in condition for allowance for the reasons that follow. Any rejections not reiterated in this action have been withdrawn. The text of those sections of Title 35, U.S. Code not included in this action can be found in a prior Office action. **This action is FINAL.**

2. Claims 50-53 and 56-61 are pending and examined in this office action.

#### ***Claim Objections***

3. Claims 60 and 61 are objected to under 37 CFR 1.75(c) as being in improper form because a multiple dependent claim because a multiple dependent claim cannot depend from another multiple dependent claim. In this case, both claims 60 and 61 depend from claim 59 which is also a multiple dependent claims. See MPEP § 608.01(n). Accordingly, the claims not been further treated on the merits.

#### ***Claim Rejections - 35 USC § 112***

4. The following is a quotation of the first paragraph of 35 U.S.C. 112:

The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

5. Claims 51, 52, 53, and 59 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claim(s) contains subject matter which

Art Unit: 1634

was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention. This is a rejection for new matter.

6. In claim 51, the limitation that the blood samples "comprises leukocytes which have not been fractionated into cell types" is new matter. Such a recitation includes, for example, testing a blood sample where the red blood cells and the white blood cells have been separated, and also includes, the testing of whole blood RNA. There is clearly basis for the latter, but not the former.

7. Applicant asserts in the remarks that this claim limitation finds clear support in the specification, including figure 5C which shows standardized fractions of leukocytes. However, these are not leukocytes that have not been fractionated into cell types, as they have clearly been fractionated into cell types. While RNA levels have been determined in each of the fractions, this is not basis for the negative limitation "have not been fractionated into cell types." There is no discussion or example in the specification of the testing of RNA in blood samples which comprise leukocytes which have not been fractionated into cell types. Applicant has attempted to present a claim which excludes a particular process step from a method (that is, fractionating the leukocytes) and then provides basis for the exclusion of the step in a method where the opposite occurred. This is not sufficient basis for the claim limitation because there is nothing in the specification that suggests applicant contemplated the exclusion of a step of fractionating leukocytes into cell types. Therefore, claims 51, as well as all claims which depend from claim 51 are rejected for having new matter.

8. Claim 59 is rejected for having new matter. Applicant did not point to basis in the specification for the negative proviso which limits the conditions which the control subjects may have.

***Claim Rejections - 35 USC § 112***

9. The following is a quotation of the second paragraph of 35 U.S.C. 112:

The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

10. Claim 59 is rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

11. It is not clear which "said control subjects" the claim refers to since the independent claim sets forth two different groups of control subjects. If applicant intends to refer to the "control subjects having osteoarthritis" it is not clear what type of osteoarthritis the subjects cannot have if they are not allowed to have mild or severe OA. The specification does not provide clear delineating factors for identifying patients with OA that is neither mild nor severe, yet is still OA, and so it is unclear which subjects would be included in this group. If applicant intends to refer to the "control subjects who are healthy" the claim seems redundant in most respects since these subjects necessarily would not have any of the recited diseases.

12. Claims 50-53 and 56-59 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the enablement requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention.

Art Unit: 1634

**Nature of the invention**

The invention is drawn to a method a method for classifying NTN4 gene expression in a human, and sets forth steps of quantifying a level of RNA encoded by a NTN4 gene, comparing that level to a level of RNA found in blood samples from control subjects having osteoarthritis (OA) and also comparing it to control subjects who are healthy. The independent claim states that based on particular determinations, the classification of NTN4 gene expression results either with that of said subjects having OA or with that of subjects who are healthy. The nature of the invention requires the knowledge of a reliable association between NTN4 expression and the ability to classify a particular individual's expression with the expression of subjects having OA or not having OA, and further, the use of this method requires that there is an underlying assumption that this classification is meaningful. Reading the claims in light of the specification it is clear that applicant intends to use such a classification method in order to provide a tool that is used as part of a diagnostic process, and such a use requires the knowledge of a reliable association underlying the classification. Further, the practice of the invention requires an understanding of how the presence of osteoarthritis effects the level of NTN4 expression in human blood.

**Scope of the claims**

The scope of the independent claim includes in step (b) patients who have OA as well as other possible conditions, including allergies, hypertension and control subject to systemic steroids. In fact, the claims are inclusive of patients who have OA plus any number of other co-morbidities such as rheumatoid arthritis or heart failure.

The claims do not recite the level of statistical significance that is required to be reached, and so, even with the requirement of statistical significance, the claims remain quite broad since no particular level is required, and the claims even encompass using different levels of statistical significance for different comparisons. The phrase “statistically significant” describes a mathematical measure of difference between groups, not a particular level of difference which is acceptable. There is no universally accepted level of “statistically significant.”

The claims remain very broad in scope because they encompass that ANY level and direction of difference in gene expression between the tested subject and the healthy controls or controls with OA is sufficient to result in a classification with the other group if the difference is “statistically significant” (provided there is also the required similarity). That is, the claims do not set forth that one level should be higher or lower than the other, and further do not set forth how much of a “difference” between two individuals would be necessary to draw the conclusions set forth in the claims.

#### **Teachings in the Specification/Examples**

Regarding osteoarthritis, the specification provides example 24 wherein gene expression profiles of blood samples from individuals having osteoarthritis were compared with normal individuals, that is healthy patients. The specification teaches that 300 genes were identified as being differentially expressed, and regarding the instant claims, table 30 provides a list of these genes (Example 24). NTN4 is among the genes.

The tables list genes that were differentially expressed, but does not provide any further information. For example, the tables do not teach if the expression was higher or lower in osteoarthritis patients versus controls.

The specification does not provide any guidance as to the level of “difference” that is sufficient (1 fold, 2 fold, etc) to result in a conclusion mRNA expression is properly classified as being in the “other” group, nor does the specification provide any guidance as to the direction of the difference (higher or lower expression) that is expected to be observed for any comparison of test subject versus the control groups. Notably, the specification also teaches that NTN4 is differentially expressed in the blood of patients who have rheumatoid arthritis versus healthy control patients (Example 22, Table 3M). In this case as well, the specification is silent as to the nature of the difference in gene expression between patients with rheumatoid arthritis and healthy controls.

Additionally, NTN4 is not reported as being differentially expressed when tests are done with patients having osteoarthritis and hypertension as compared with normal patients (Table 3A), or osteoarthritis and obesity as compared with normal patients (Table 3B) or osteoarthritis and allergies as compared with normal patients (Table 3C), or osteoarthritis and subject to systemic steroids compared with normal patients (Table 3D). Furthermore, the gene does not appear in Table 3AB which provides genes that are differentially expressed in blood from patients with mild or severe OA but genes relevant to asthma, obesity, hypertension, systemic steroids and allergies have been removed. Thus, the gene is not differentially expressed in all patients with osteoarthritis.



Art Unit: 1634

The specification fails to provide information about an essential aspect of the invention, namely, the nature of the difference in expression that was observed between osteoarthritis patients and healthy patients. This information is essential to understanding and practicing the claimed invention because it is critical to knowing how to interpret a particular comparison result. Although the claims are drawn to a method of "classifying expression," and not to diagnosis or identifying increased likelihood of disease or the like, it is critical to understand how the classification can be used in order use the claimed invention. In this case, the specificaiton does not provide sufficient guidance as to how to use the classification method, or in other words what is the meaning of classifying expression "with that of subjects having" OA or with subjects who are healthy? While one could do the method steps as written, thus satisfying the "how to make" aspect of 112 1st paragraph, the specification does not provide sufficient disclosure to satisfy the how to use aspect of the requirement.

**State of the Prior Art and Level of Unpredictability**

Observing differences in expression between two populations is a highly unpredictable endeavor. The specification clearly exemplifies this for the case of NTN4. The specification teaches differential expression of this gene between populations of patients with osteoarthritis and healthy controls. The specification also teaches that NTN4 is differentially expressed in the blood of patients who have rheumatoid arthritis versus healthy control patients (Example 22, Table 3M). So first, even if one carried out the claimed analysis on a test subject, and if one observed a level of expression, it is highly unpredictable how would one begin to know if that level of expression would properly be classified with patients having osteoarthritis, rheumatoid

Art Unit: 1634

arthritis, both, one but not the other, something in between or even some other condition or disorder for which the expression profile has not yet been determined.

Further, NTN4 it is not listed in the tables for differentially expressed genes in patients who have both osteoarthritis and other diseases or conditions versus normal controls. So, this exemplifies that a particular gene is not always differentially expressed in populations of patients having osteoarthritis versus healthy controls, and it also suggests that NTN4 expression could often be classified, according to the claimed methods, as being "with that of said subjects who are healthy" when the subject actually has OA. Observing the differential expression result is population dependent- something about patients with osteoarthritis and other diseases or conditions changes the observation. It is unknown and unpredictable whether this is also true for differential expression observations in other diseases. Furthermore, although NTN4 was not observed to be differentially expressed in any of the other examples in this specification, it is unknown and unpredictable whether it would be expressed in the blood of patients having other bone related diseases or other types of arthritis or any other diseases which were not tested in the instant specification or diseases which were tested in the instant specification but in a different population of test subjects, and whether this expression would be different from levels of expression in healthy controls. A method which relies on a comparison between expression in the blood of a test subject and control subjects requires the knowledge of this information in order to understand why it is relevant that expression of a gene in an individual is classified with subjects having OA or healthy subjects, as set forth in the claims. The instant specification has not established that all difference, no matter the magnitude nor the direction, relative to any control subjects or even relative to a healthy control subject is related to osteoarthritis (which is,

Art Unit: 1634

of course, what is suggested by classifying gene expression in said test subject with said subjects having OA). In fact, the specification shows that RA patients have a difference in this gene relative to healthy controls. It is not known under what circumstances the result observed in the instantly examined control and test populations would be repeatable, as the results have not been validated. But even if one were to obtain the same result, it would be unknown because applicant did not disclose the magnitude of difference in expression between osteoarthritis patients or controls, nor did applicant disclose the direction of variation. All of these inquiries are particularly important in this case since the specification is silent as to which differential expression observations would be sufficient understand how to use (i.e. apply) a method which classifies gene expression of NTN4 with OA patients or with healthy subjects.

Because the claims encompass any level of altered gene expression, it is relevant to point out that the art of Cheung et al (2003) teaches that there is natural variation in gene expression among different individuals. The reference teaches an assessment of natural variation of gene expression in lymphoblastoid cells in humans, and analyzes the variation of expression data among individuals and within individuals (replicates) (p.422, last paragraph; Fig 1). The data indicates that, for example, expression of ACTG2 in 35 individuals varied by a factor of 17; and that in expression of the 40 genes with the highest variance ratios, the highest and lowest values differed by a factor of 2.4 or greater (Fig 3). It is thus unpredictable as to whether or not any level of altered gene expression is indicative of a osteoarthritis or the absence of osteoarthritis cancer.

The unpredictability of correlating gene expression level to any phenotypic quality is taught in the post-filing art of Wu (2001). Wu teaches that gene expression data, such as

Art Unit: 1634

microarray data, must be interpreted in the context of other biological knowledge, involving various types of 'post genomics' informatics, including gene networks, gene pathways, and gene ontologies (p.53, left col.). The reference indicates that many factors may be influential to the outcome of data analysis, and teaches that expression data can be interpreted in many ways. The conclusions that can be drawn from a given set of data depend heavily on the particular choice of data analysis. Much of the data analysis depends on such low-level considerations as normalization and such basic assumptions as normality (p.63 - Discussion). The art of Newton et al (2001) further teaches the difficulty in applying gene expression results. Newton et al. teaches that a basic statistical problem is determining when the measured differential expression is likely to reflect a real biological shift in gene expression, and replication of data is critical to validation (p.38, third full paragraph). There is no replication of data in the instant specification.

### **Quantity of Experimentation**

The instant specification does not provide enabling support for the practice of a single embodiment within the claimed invention. In particular, the specification does not provide adequate guidance to appraise one of ordinary skill in the art as to what levels of NTN4 gene expression must be observed for the conclusions set forth in the claims to be meaningful. Further, although the specification teaches there are differences in NTN4 levels in a osteoarthritis population versus a control patient population, the specification is silent as to the nature of the "difference" in magnitude or direction. Thus, given the lack of teaching in the specification and the highly unpredictable nature of the technology, an extensive amount of work would be required to practice the claimed invention.

Art Unit: 1634

In order to practice the claimed invention, one would have to undertake an extensive amount of experimentation in a highly unpredictable technology area. One would begin by trying to reproduce the results observed in the instant specification to determine if there is a relative upregulation or downregulation of NTN4 in osteoarthritis patients versus healthy control patients, as the specification does not even provide this minimal guidance. Without this knowledge one would not even begin to know how to interpret any results obtained in practicing the claimed methods. For example, consider the comparison of a test result and a control population of healthy individuals. How different from the average level of expression of healthy individuals would the test result have to be to indicate osteoarthritis? Would any difference, up or down regulation be indicative of osteoarthritis? Or could one indicate osteoarthritis and one rheumatoid arthritis? Is NTN4 expressed in the blood of individuals with a disease other than rheumatoid arthritis and osteoarthritis? Is this expression also diagnostic of other degenerative diseases or other diseases of the joints or other disorders entirely unrelated to osteoarthritis? What populations could be tested and expected to provide a reliable result, as it is clear from the results in the specification that not all control subjects having osteoarthritis differentially express NTN4 relative to healthy controls? For example, if a subject has allergies, would the practice of the claimed assay be reliably informative in the event that NTN4 expression is different or the same as the control subjects? In order to reliably use a method for detecting osteoarthritis, one would first have to answer at least these questions. One would also, however, have to carry out this testing for validation, for it is possible that the result observed in the instant specification is intrinsic to the cohort of patients evaluated in applicant's study. Further, one would have to

Art Unit: 1634

undertake experimentation to determine difference thresholds required to determine that a patient has or does not have a disease.

As discussed, this art area is highly unpredictable.

### **Conclusion**

The claims include methods which encompass the detection in blood of the expression of NTN4 in a test subject and comparing this expression to control subjects, wherein the results are used to “classify the expression” as being with OA subjects or with healthy subjects. The identification of gene differential expression/disease indication relationships is a highly unpredictable endeavor, requiring extensive experimentation. The specification provides minimal guidance. In light of the factors discussed, therefore, it is concluded that it would require undue experimentation to practice the claimed invention.

### **Response to Remarks**

Applicant traverses the rejection for lack of enablement, on page 12 stating that since the claims are not drawn to a method of diagnosis but merely to classifying gene expression the instant claims are enabled (p. 12). Further, applicants point out that NTN4 was shown to be differentially expressed between subjects having OA and healthy controls (p. 13). However, this is not persuasive, as discussed in the rejection which has been modified to address the newly added claim. First, the specification does not provide sufficient guidance or information to even support that the classification as set forth makes sense or is valid, let alone to appraise one of skill in the art as to how to use the invention if one were to practice it. Since NTN4 is taught to be differentially expressed in the experiments with OA patients and RA, the specification does not provide guidance as to how to use a method which classifies the expression as set forth in the

Art Unit: 1634

claims. Further, given that many examples which include OA patients DID NOT find a difference in expression relative to healthy patients, it seems equally likely that a test subject would be misclassified. The specification demonstrates that the potential relationship between NTN4 expression and OA is highly unpredictable and population specific, yet provides no clear guidance as to when one could rely on a particular result. One would not know, based on the teachings of the specification how or if the classifying result of the claimed invention is meaningful.

Also, the current claims do not require a particular level of “statistical significance” and so this aspect of the claims is still quite broad, and in fact a different level could be required for each comparison in the claims.

On page 14 applicant argues that the instant claims are “sufficiently predictable for enablement” because they merely require determination of statistical similarity and difference in gene expression levels. As previously noted in this office action, however, it is not predictable how to use such a method, that is what the possible meaning of such a finding might be, or even that such a finding would have robust implications.

Applicant states that the results of Cheung et al. cannot be reliably extrapolated to primary blood samples since Cheung et al. are using cultured cell lines. However, this is irrelevant to the point of Cheung et al. which is that among individuals (in this case cell lines) there is natural variability in gene expression for any particular gene. Attorney arguments are not sufficient to establish that this biological fact is not the case.

Applicant disagrees with Wu that expression data needs to be interpreted in view of other biological knowledge. Wu was relied upon for much more than this simple statement. Wu

Art Unit: 1634

discusses at length many of the factors that make gene expression analysis unpredictable.

Applicant's statement that "differential gene expression which is reproducible, and is correlated with the state of health or disease of the individual does not necessarily result directly in the state of the disease of the individual" is attorney argument which is not supported by evidence on the record. Even if the changes are a result of downstream effects of the pathogenic process, they are related to the state of disease in the individual. Applicant points out that certain prostate markers were used as biomarkers without an understanding of their function. The examiner is not trying to require an understanding of NTN4 in OA or any other disease, nor does Wu suggest that such is necessary. The examiner is looking to the specification for adequate guidance for making and using an invention in a highly unpredictable field of endeavor.

### *Conclusion*

13. No claim is allowed.

14. Applicant's amendment necessitated the new ground(s) of rejection presented in this Office action. Accordingly, **THIS ACTION IS MADE FINAL**. See MPEP § 706.07(a).

Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event,



Art Unit: 1634

however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

15. Any inquiry concerning this communication or earlier communications from the examiner should be directed to Juliet C Switzer whose telephone number is (571) 272-0753. The examiner can normally be reached on Monday, Tuesday, or Thursday, from 9:00 AM until 4:30 PM.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Ram Shukla can be reached by calling (571) 272-0735.

The fax phone numbers for the organization where this application or proceeding is assigned are (571) 273-8300. Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is (571)272-0507.

Patent applicants with problems or questions regarding electronic images that can be viewed in the Patent Application Information Retrieval system (PAIR) can now contact the USPTO's Patent Electronic Business Center (Patent EBC) for assistance. Representatives are available to answer your questions daily from 6 am to midnight (EST). The toll free number is (866) 217-9197. When calling please have your application serial or patent number, the type of document you are having an image problem with, the number of pages and the specific nature of the problem. The Patent Electronic Business Center will notify applicants of the resolution of the problem within 5-7 business days. Applicants can also check PAIR to confirm that the

Art Unit: 1634

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For all other customer support, please call the USPTO Call Center (UCC) at 800-786-9199.

/Juliet C. Switzer/  
Primary Examiner  
Art Unit 1634

December 12, 2007